July 31, 2002

Anne P. LeHuray, Ph.D.
Monocyclic Aromatic Amines and Nitroaromatics Panel
American Chemistry Council
1300 Wilson Boulevard
Arlington, VA 22209

Dear Dr. LeHuray:

The Office of Pollution and Toxics is transmitting EPA's comments on the robust summaries and test plan for m-Nitrotoluene, posted on the ChemRTK HPV Challenge Program Web site on December 7, 2001. I commend the American Chemistry Council's Monocyclic Aromatic Amines and Nitroaromatics Panel for its commitment to the HPV Challenge Program.

EPA reviews test plans and robust summaries to determine whether the reported data and test plans will provide the data necessary to adequately characterize each SIDS endpoint. On its Challenge Web site, EPA has provided guidance for determining the adequacy of data and preparing test plans used to prioritize chemicals for further work.

EPA will post this letter and the enclosed Comments on the HPV Challenge Web site within the next few days. As noted in the comments, we ask that the Panel advise the Agency, within 90 days of this posting on the Web site, of any modifications to its submission.

If you have any questions about this response, please contact Richard Hefter, Chief of the HPV Chemicals Branch, at 202-564-7649. Submit questions about the HPV Challenge Program through the HPV Challenge Program Web site "Submit Technical Questions" button or through the TSCA Assistance Information Service (TSCA Hotline) at (202) 554-1404. The TSCA Hotline can also be reached by e-mail at tsca-hotline@epa.gov.

I thank you for your submission and look forward to your continued participation in the HPV Challenge Program.

Sincerely,

Oscar Hernandez, Director
Risk Assessment Division

Enclosure

cc: W. Sanders
A. Abramson
C. Auer
M. E. Weber

**EPA Comments on Chemical RTK HPV Challenge Submission:**
**m-Nitrotoluene**

**SUMMARY OF EPA COMMENTS**

The sponsor, the Monocyclic Aromatic Amines and Nitroaromatics (MAANA) Panel of the American Chemistry Council, submitted a Test Plan and Robust Summaries to EPA, dated November 13, 2001, for m-Nitrotoluene (CAS No. 99-08-1). EPA posted the submission on the ChemRTK HPV Challenge Web site on December 7, 2001.

EPA has reviewed this submission and has reached the following conclusions:

1. Data adequacy. Most of the robust summaries need revision to be acceptable for the Challenge Program. EPA has provided specific comments on how to enhance the robust summaries to the standard established in EPA's HPV Challenge Program Guidance (http://www.epa.gov/opptintr/chemrtk/guidocs.htm).

2. Physicochemical and Environmental Fate Data. The submitter needs to address stability in water (hydrolysis) in the test plan. The submitter needs to recheck the data and references for hydrolysis in the robust summary. The submitter needs to re-evaluate the ready biodegradation data.

3. Health Effects. EPA reserves judgement concerning the adequacy of the available data for purposes of the HPV Challenge Program for acute, repeated-dose, and genetic toxicity endpoints pending submission of necessary missing data elements in the robust summaries. EPA considers that the reproductive and developmental toxicity endpoints have not been adequately addressed and recommends conducting a combined reproductive/developmental toxicity screening test to address these endpoints, because effects on male and female reproductive organs were seen in the NTP repeated-dose toxicity study.

4. Ecotoxicity. EPA considers the existing data to be adequate for the fish endpoint only. EPA reserves judgement on the adequacy of the invertebrate and algal studies pending submission of necessary missing data elements in the robust summaries.

EPA requests that the submitter advise the Agency within 90 days of any modifications to its submission.

**EPA COMMENTS ON m-NITROTOLUENE CHALLENGE SUBMISSION**

**Test Plan**

Chemistry (melting point, boiling point, vapor pressure, water solubility, and partition coefficient).

The submitter's approach to the chemistry endpoints is acceptable for the purposes of the HPV Challenge Program.

Environmental Fate (Photodegradation, Stability in Water, Biodegradation, Fugacity).

*Stability in Water*. Although this substance has no hydrolyzable functions, the robust summary shows18% degradation after 8 days at pH 7.4 and 25 °C. The chemical's photodegradation half-life of 2.6 hours suggests that the apparent degradation could be a result of photolysis. However, the submitter failed to

address this situation. The submitter needs to discuss the unexpected results and consider whether any further testing may be necessary to resolve the issue.

*Biodegradation*. The submitter ran two ready biodegradability tests, one for 14 days (OECD 301 C) and the other for 20 days (OECD 301 D). The robust summaries suggest that the two biodegradability tests may have met the guideline's criteria for termination prior to 28 days, but the criteria for ready biodegradability were not met. However, EPA believes that this substance may be readily biodegradable. Therefore, unless it can be demonstrated that ending the existing tests before 28 days was appropriate *because* pass levels were reached, the submitter needs to provide ready biodegradability data from a 28-day test.

Health Effects (acute toxicity, repeated-dose toxicity, genetic toxicity, and reproductive/developmental toxicity).

EPA reserves judgement concerning the adequacy of the available data for purposes of the HPV Challenge Program for acute, repeated-dose, and genetic toxicity endpoints pending submission of necessary missing data elements in the robust summaries. EPA considers that the reproductive and developmental toxicity endpoints have not been adequately addressed and recommends conducting a combined reproductive/developmental toxicity screening test to address these endpoints, because effects on male and female reproductive organs were seen in the NTP repeated-dose toxicity study.
*Acute toxicity.* Although the adequacy of individual acute oral toxicity studies could not be determined from the limited information provided, the studies consistently report $LD_{50}$ values (1070-2200 mg/kg) that approach or equal the value for a limit test under OECD guidelines. Pending submission of revised robust summaries, the data available for the acute oral toxicity endpoint appear to be adequate for the purposes of the HPV Challenge Program.

Page 2 of the test plan states that the range for oral $LD_{50}$ values in rats is >500 to <2000 mg/kg, which is not in agreement with data in the robust summaries: range 1070-2200 mg/kg. The submitter needs to address this discrepancy.

*Reproductive and Developmental Toxicity*. EPA considers the critical study cited for the reproductive and developmental toxicity endpoints (Ciss 1978, Ciss et al. 1980) inadequate to address these endpoints, since the protocol is not a standard study, only one dose level (300 mg/kg/day) was tested, group sizes were small, and no effects were seen on either reproductive or developmental parameters. The repeated-dose toxicity (GLP) study conducted at NTP (Dunnick 1992, Dunnick et al. 1994), however, showed testicular degeneration, and decreased epididymal sperm count and concentration at 750 mg/kg/day in male rats. Increased female estrous cycle length and increased number of affected (non-cycling) females were also seen at 375 and 750 mg/kg/day. Therefore, EPA recommends that the submitter conduct a combined screening test for reproductive and developmental toxicity using OECD Guideline 421.

Ecotoxicity.

Except for adequate data on one fish study, the robust summaries lack sufficient data to assess the studies, and EPA will defer judgement on the invertebrate and algal toxicity endpoints until the data are provided.

**Specific Comments on the Robust Summaries**

General. The submitter needs to provide the identity and purity of the test material for each test described, or indicate where such data are not available from the original report.

Since the GLP guidelines were not in existence prior to 1980, the submitter needs to change the notation in the robust summaries from “no data” to “no” for the older studies.

Environmental Fate.

*Stability in Water*. The robust summary lacks important details; for example, data are given for only one pH value. According to OECD guideline 111, hydrolysis half-lives should be determined over a range of environmental pH values (pH 4,7 and 9); only if <10 % hydrolysis is seen after 5 days (at 50 °C) can the test substance be considered to be hydrolytically stable and no further testing be necessary. Finally, the reference for the test method appears to be incorrect.

Health Effects.

All robust summaries are incomplete. For the specific omissions outlined below, the submitter needs to indicate which data elements were not reported in the original study or add the available reported data to the robust summary. Summaries of all animal studies need to include the strain and sex, if known.

*Acute Toxicity*. The robust summary for the critical study (Ciss et al. 1980, Ciss 1978) in rats needs to include the administered dose levels, the number of animals per sex per dose, the length of the observation period, the results for mortality and toxicity by dose and sex, statistical methods, and any clinical observations or non-lethal toxic effects. The submitter needs to describe the experimental method in the robust summary, rather than only providing a reference to a published method. If sufficient information to evaluate the study cannot be reported, the submitter needs to report additional details for the supporting studies for evaluation by EPA.

*Repeated-Dose Toxicity*. The submitter needs to add to the critical NTP Program 13-week studies in rats and mice the year of the study, the frequency of data collection for clinical signs, the specific hematological and clinical chemistry endpoints evaluated, the specific organs/tissues that were weighed and examined histologically, including reproductive organs, and the statistical methods used.

*Genetic Toxicity*. The submitter needs to add to the robust summary for the critical study on reverse mutation in bacteria the chemical name and purity of the test substance (since section 1.1-1.4 of the dossier is empty), the positive controls, and the number of replicate plates. For the critical study on chromosomal aberration in Chinese hamster ovary (CHO) cells, the submitter needs to add the purity of the test substance, the number of metaphases that were evaluated, the source of the metabolic activation system, and the positive controls. This same study appears to be duplicated as a supporting study on page 40/55; the submitter needs to delete this second entry if it is a duplicate.

Ecotoxicity.

For the algal and invertebrate toxicity robust summaries, the submitter needs to add the missing information such as species, test duration, test type (static vs. renewal), endpoint value, concentrations tested, number of daphnids/replicates tested, water chemistry details, control data, test substance purity, cumulative mortality, $LC_{50}$ value, and statistical methods used.

**Follow-up Activity**

EPA requests that the submitter advise the Agency within 90 days of any modifications to its submission.